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14. ABSTRACT The high incidence of mild TBI in Warfighters exposed to blast has triggered interest in injury mitigation and increased TBI resilience as well as concerns over safe return-to-duty and minimization of long-term and delayed TBI-related debilitations in returning veterans. These objectives require the utilization of high fidelity animal models to investigate the underlying neurobiological mechanisms of injury as a rational basis for defining risks and establishing effective countermeasures. The etiology of blast TBI (bTBI) is largely undefined, and several mechanisms, likely interactive, have been proposed. Using a well-validated blast model, we are exploring blast-induced acceleration of the head as one of the primary components of bTBI. Having successfully worked with a range of inanimate objects of varied areal densities to define relations of blast flow conditions to acceleration and displacement, we have begun examination of the effects of systematically varied conditions on anesthetized rats to isolate and distinguish the contributions of blast-induced head acceleration and displacement from other biomechanical components and effects of the shockwave.					
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Table of Contents

	<u>Page</u>
Introduction.....	3
Body.....	4
Key Research Accomplishments.....	7
Reportable Outcomes.....	7
Conclusion.....	7
References.....	8
Appendices.....	8

INTRODUCTION: The prevalence of blast-induced TBI (bTBI) has prompted an urgent need to develop improved mitigation strategies and advance medical care targeting casualties with bTBI. Despite considerable effort and a broadened interest in the study of mild TBI of all causes, the basic mechanisms of blast induced brain injury are for the most part still undefined. Based largely upon computational modeling and the parallel evolution of interest in athletic concussive head injuries, several candidate mechanisms of non-impact blast TBI have been identified and include head acceleration. Animal models are required to verify and validate these models, to identify the underlying biomechanical and neurobiological events resulting in injury, and to establish effective countermeasures. Examination of each of these proposed mechanisms requires shock wave exposure conditions and specimen targets that are appropriate for the question being asked. We believe that the rat can be employed in a laboratory simulation of blast in a manner that directly addresses the role of acceleration as a critical component of bTBI.

We hypothesize that explosion flow conditions can cause head acceleration sufficient to injure the brain, and that these inertial forces combine with other injury mechanisms to yield blast TBI. Based upon empirically defined relations between blast flow conditions (e.g. peak static and total pressure, positive phase duration, and impulse) and acceleration and displacement of a wide range of inanimate objects, we have begun examination of the effects of systematically varied conditions on surrogate rats, cadaveric rat specimens, and anesthetized rats to isolate and distinguish the contributions of blast-induced head acceleration and displacement from other biomechanical components and effects of the shockwave.

Using a highly characterized simulation of blast in an Advanced Blast Simulator (ABS) in the laboratory, rats are exposed to BOP with varied peak amplitudes and impulse in comparison with systematically matched and carefully controlled independent acceleration of head and torso. High speed video recordings of experimental subjects during blast exposure and independent acceleration are closely compared and the resultant pressure responses in varied compartments in concert with the neuropathological, neurochemical, and neurobehavioral consequences of exposures under these conditions provides a basis for determination and quantitation of the underpinnings of blast TBI.

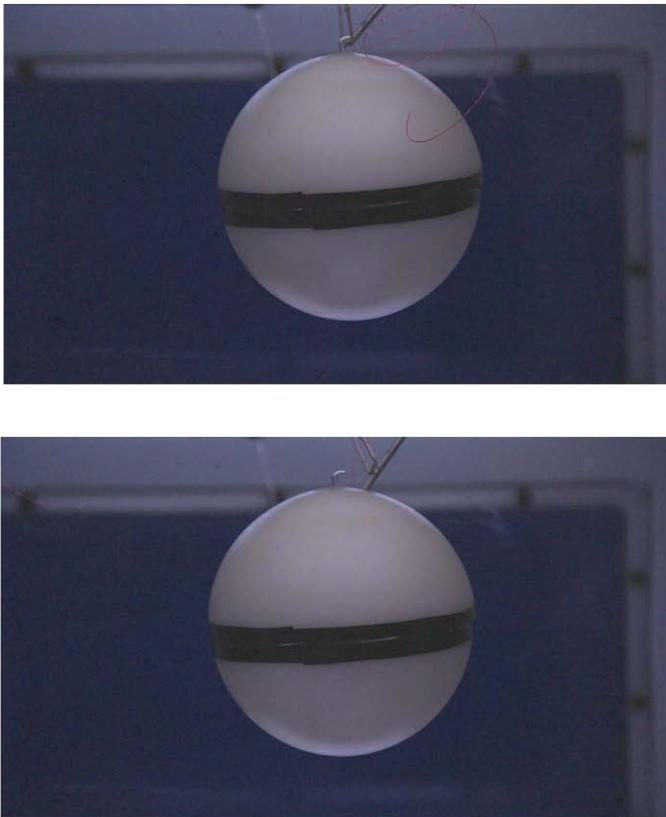
BODY: A primary innovation of this project is the continued development, characterization, and utilization of high fidelity simulations of blast flow conditions in an Advanced Blast Simulator that are possible through close consultation with blast physics experts who are actively involved in this project. With this device it is possible to replicate all key features of blast flow wave conditions, including the negative phase and secondary shock. Analysis and tight control of these components (notably acceleration and displacement) in combination with functional outcome measures provides a means to understand the relation of the former to the latter. An explosive shockwave is unlike any other conventional mode of loading and imparts both an abrupt transient crushing action (i.e. static pressure) which envelops the head as well as some aerodynamic drag (i.e. dynamic pressure creating blast wind). The use of animal models to investigate blast-induced neurotrauma requires appreciation of the relative biomechanics and scaling; it is essential to replicate the proper incident blast conditions to assess relevant brain injury mechanisms. Ongoing controversies and confusion concerning the contributions of blast-induced head acceleration to blast-induced TBI have in great part resulted from laboratory studies in which blast was inappropriately simulated, and head acceleration was likely in many cases an experimental artefact uniquely associated with those particular exposure conditions. Experiments with an advanced blast simulator (ABS) yield a higher fidelity, ecologically valid simulation of blast and thereby provide critical insights into the etiology of

bTBI that can serve to guide the rational development of mitigation measures and further elucidate pathophysiological mechanisms that can be therapeutically targeted.

The project has been approached in 3 stages. During the first stage, an adult-size and weight surrogate rat and rat cadaveric specimens have been used along with other appropriately scaled inanimate objects to record acceleration and displacement resulting from BOP and to establish the operational parameters required to evaluate relations between pressure conditions (i.e. peak pressure and impulse) and resultant acceleration and displacement. In the second stage, under these well-defined exposure conditions, anesthetized rats are used to simultaneously record intracranial pressure (ICP), intravascular pressure, and acceleration/displacement of the head and trunk. Tissue samples are used to investigate the neurobiological underpinnings of the brain injuries resulting from these blast-induced biomechanical conditions. In the third stage, rats subjected to each of these injury conditions (blast and acceleration vs acceleration alone) undergo neurobehavioral and histopathological assessments to comprehensively characterize and compare the resultant injuries and functional impairments.

Task 1. Using rat surrogates in a 24 in diameter advanced blast simulator (ABS), determine the exposure parameters required to optimally and independently manipulate pressure conditions (i.e. peak pressure and impulse) and acceleration and displacement. Establish 12 exposure conditions (including controls) that will be used to systematically pair BOP peak pressure/impulse (3 intensities) and acceleration/displacement (3 intensities).

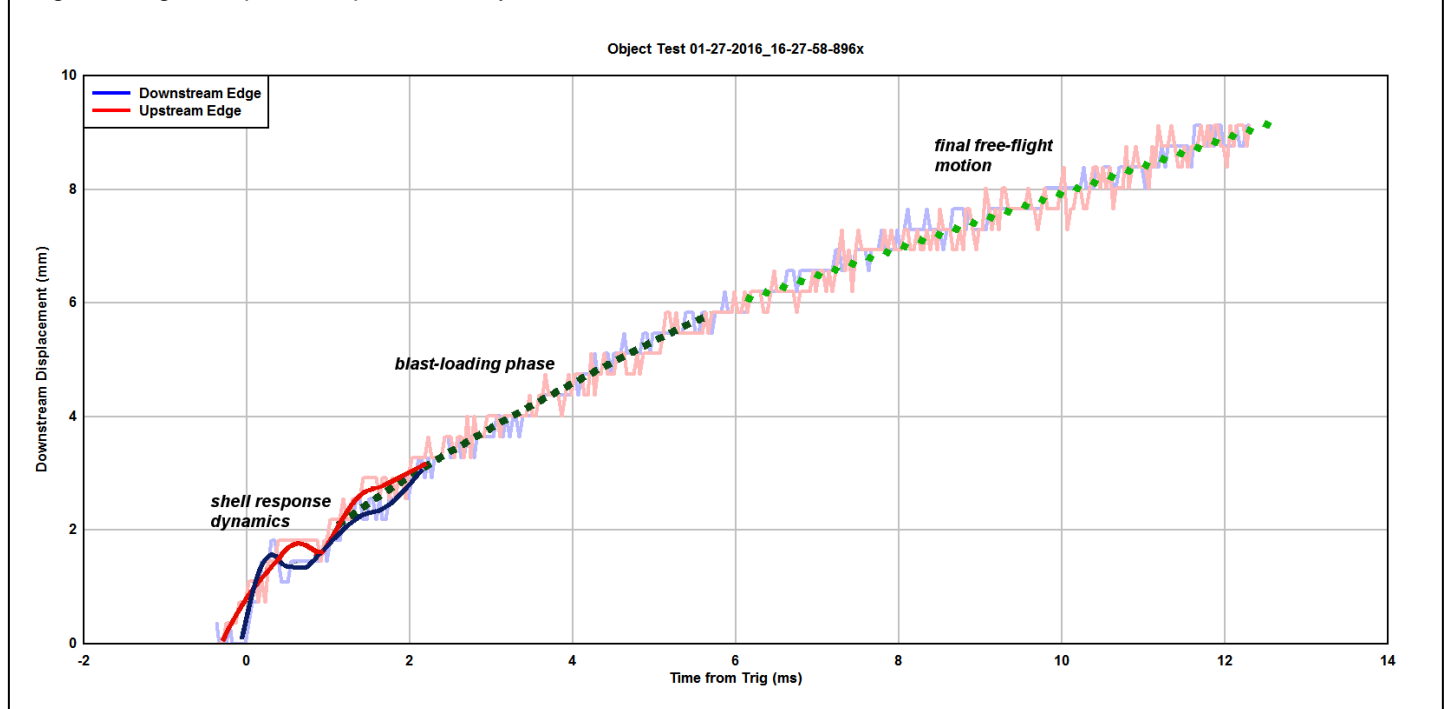
Fig 1. Weighted sphere release mechanism. The restraining rod is pulled away just before the shock wave arrives - note the lack of movement of red filament. (top). Release mechanism 140 msec after arrival of blast (bottom).



During this reporting period, the test section of the ABS was modified to incorporate enlarged Plexiglas windows to allow full flight high speed video recordings of blast exposed targets. Viewing ports were enlarged from 6 inch x 6 inch to 18 inch x 12 inch. In addition, a state-of-the art high speed video camera was procured to provide the combined resolution (702 x 1280 pixels) and speed (25,000 frames per sec) to capture and analyze high speed events. The combination of speed and resolution allows researchers to visualize and calculate the forces as the shock wave initially interacts with objects. Characterization and range-finding experiments were continued with the ABS to define and refine the optimal means to create and monitor varied flow conditions and to explore unrestrained displacement and acceleration of test specimens under these varied flow conditions. In particular, to extend earlier experiments systematically determining the contribution of the shock diffraction phase versus the loading phase of the shock wave

to the ultimate velocity imparted to an object, a set of 16 specially weighted spheres (consisting of 4 different weights and 4 different diameters) were prepared and a custom release mechanism using a spring and retaining bar system, somewhat like a mouse trap, was created to release the balls just before the shock wave impacts the ball. A release mechanism that does not produce any external forces is crucial in determining the nature of the interaction of the spheres and the shock wave. In ongoing evaluations to identify the optimal frangible material to separate driver and test sections, we recognized substantial improvements with the architectural fabric manufactured by Mehler Technologies (GmbH) called Valmex. This polyvinylidene fluoride material contains a proprietary fabric weave in the center which produces excellent strength and allows evaluations with greater pressures than could be achieved to date. A single .034" thick Valmex membrane produces a blast static pressure of 20 psi. To achieve that pressure with the previously-used acetate/fiberglass screen combination membrane would require 2 pieces of acetate and 3 separate pieces of screen mesh. In addition, unlike the acetate and screen mesh membranes, Valmex does not shatter into tiny fragments, but rather rips down the middle, eliminating potential complications associated with flying membrane debris. A final critical accomplishment during this reporting period was fashioning a means to secure rats in the ABS for BOP exposure without constraining them and potentially interfering with blast-induced acceleration. After evaluation of various approaches, a linear bearing

Fig 2. Weighted sphere displacement by BOP in ABS.



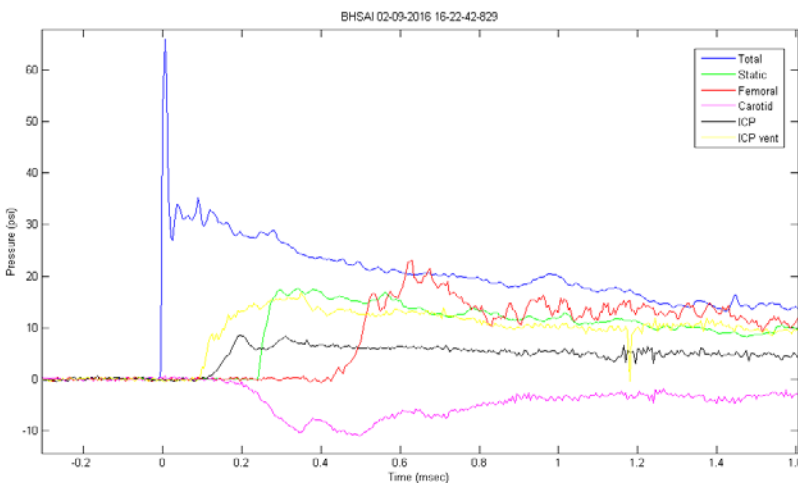
slide was developed that nicely serves this purpose.

Task 2. Using the systematic pairings of exposure conditions defined in task 1, record head and trunk acceleration and intravascular and ICP responses under each BOP exposure condition. Collect blood and brain tissue 24 h after exposures to establish neurochemical and biomarker correlates.

Progress toward completion of Task 2 during this reporting period includes the review and approval

of an animal use protocol by the WRAIR/NMRC IACUC and the ACURO. With the animal use protocol in place, as illustrated in figures 3 & 4 below, substantial progress was made to record acceleration/displacement of the experimental subjects under different flow conditions and compare ambient pressures with those recorded in various anatomical locations in the rat.

Fig 3. Ambient (total and static) and intracompartmental pressure recordings during BOP exposure. ICP recordings were made peridurally and intraventricularly.



Task 3. Using the systematic pairings of exposure conditions defined in task 1, record accelerations and identify the ensuing neurobehavioral disruptions and histopathological consequences resulting from each of the 12 exposure conditions.

Neurobehavioral and histopathological assessments were not performed during this reporting period.

KEY RESEARCH ACCOMPLISHMENTS:

The experimental means to reproducibly generate shock waveforms of varied amplitudes and durations in the ABS were carefully refined. The means to secure test objects and experimental subjects without constraining acceleration resulting from blast were developed and applied. High speed high resolution videography was greatly improved and employed to define blast-induced motion and establish scaling for model assessments of rat TBI. These results with inanimate test subjects importantly revealed that displacement is dominated by the diffraction phase of the shock wave rather than by the dynamic pressure impulse or blast wind and is now being extended to recordings with living specimens. Based upon these data, an acceleration test device has been designed and is under construction, and will provide a means to isolate and distinguish the contributions of acceleration to the brain injuries resulting from blast.

REPORTABLE OUTCOMES: None at this point

CONCLUSION: Despite widespread recognition of the urgent need to develop improved mitigation strategies and advance medical care targeting casualties with blast-induced traumatic brain injury (bTBI), the basic mechanisms of blast induced brain injury are for the most part still undefined or misidentified due to the widespread use of experimental simulations of blast that fail to account for blast flow conditions and the biomechanical loading conditions yielding bTBI among injured Warfighters. Examination of mechanisms and identification of countermeasures require shock wave exposure conditions and specimen targets that are appropriate for the question being asked. By

incorporating blast physics expertise into biomedical research, in this project we are replicating all key features of blast flow wave conditions and directly addressing the contribution of blast induced acceleration and displacement to brain injuries and disrupted functional outcome.

REFERENCES: None

APPENDICES: None

SUPPORTING DATA:

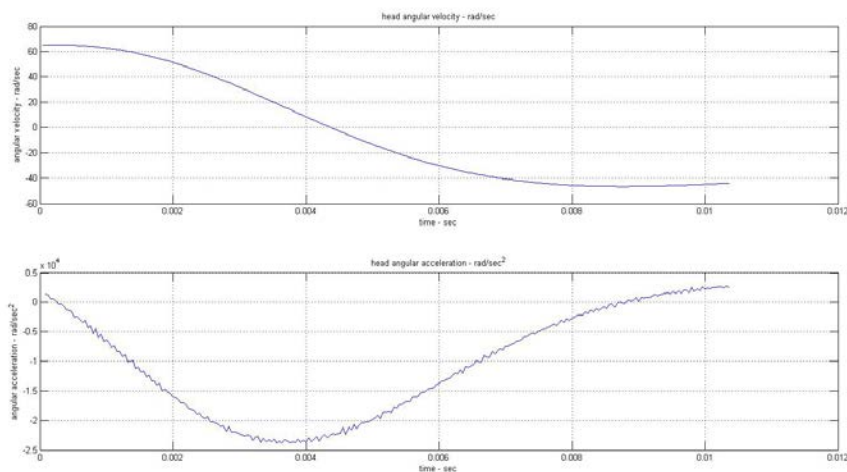
Target velocity and acceleration curves:

Blast can create injuries by a number of mechanisms. Direct injuries from blast waves interacting with the skull and brain, blast induced acceleration injuries, and impact injuries are three of the more common suspected mechanisms. A primary goal of this project is to evaluate blast induced acceleration as a potential injury mechanism. To accomplish this, targets are exposed to actual blast in the 24 inch ABS; both direct blast wave and acceleration effects will result from these exposures. The resulting injuries are then compared to the injuries resulting from exposure of similar experimental subjects to comparable acceleration but without the direct blast wave effects of the actual blast exposures. To accomplish this, a device must be designed and constructed that will accelerate targets with both rotational and linear components comparable to that resulting from actual blast exposure. This device will be used to induce acceleration injuries, if any, to allow the comparison to the blast induced injuries.

A comprehensive survey of the literature indicated there was no previous work that quantified the linear and angular acceleration of rats resulting from blast exposures comparable to that which can be achieved in the WRAIR ABS. This information is necessary to properly design the acceleration device.

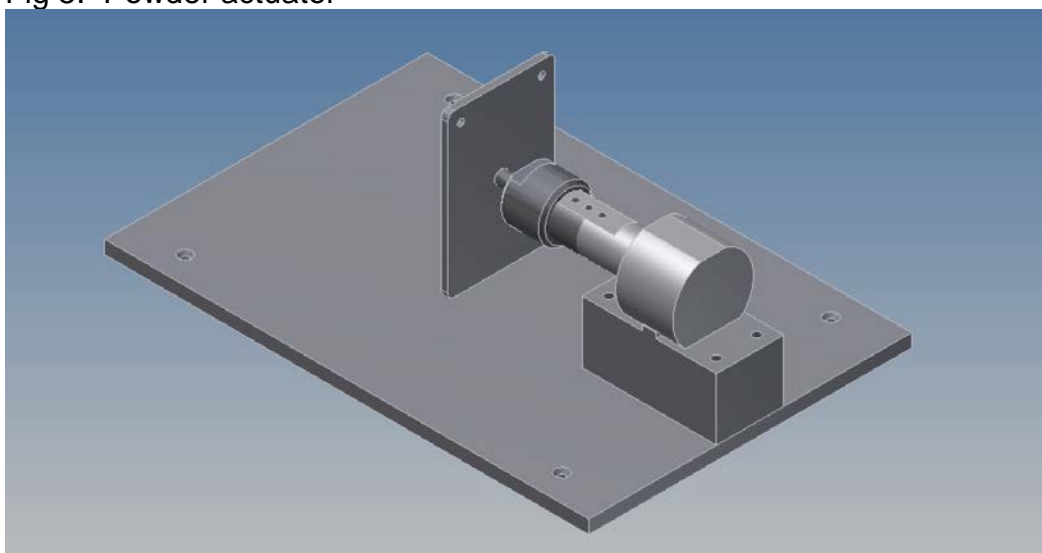
During this reporting period, researchers at WRAIR conducted a series of blast exposures of rat surrogates and rat cadaveric specimens to collect video records for analysis. These records were segmented into individual frames and each frame was analyzed using a point to point tracking algorithm to determine the acceleration in the sagittal plane. These results indicated that the target was fully accelerated very early following the arrival of the shock wave. Using video recorded at 25,000 frames per second, the results indicated that the target was fully accelerated by the third

Fig 4: Angular velocity and acceleration of the rat head during blast exposure



frame (0.12 milliseconds) after shock wave arrival. The blast waves had a positive phase of approximately 5 milliseconds. From these results it is determined that the target is fully accelerated in the diffraction phase of the target-shock interaction and there is no significant drag phase acceleration.

Fig 5: Powder actuator



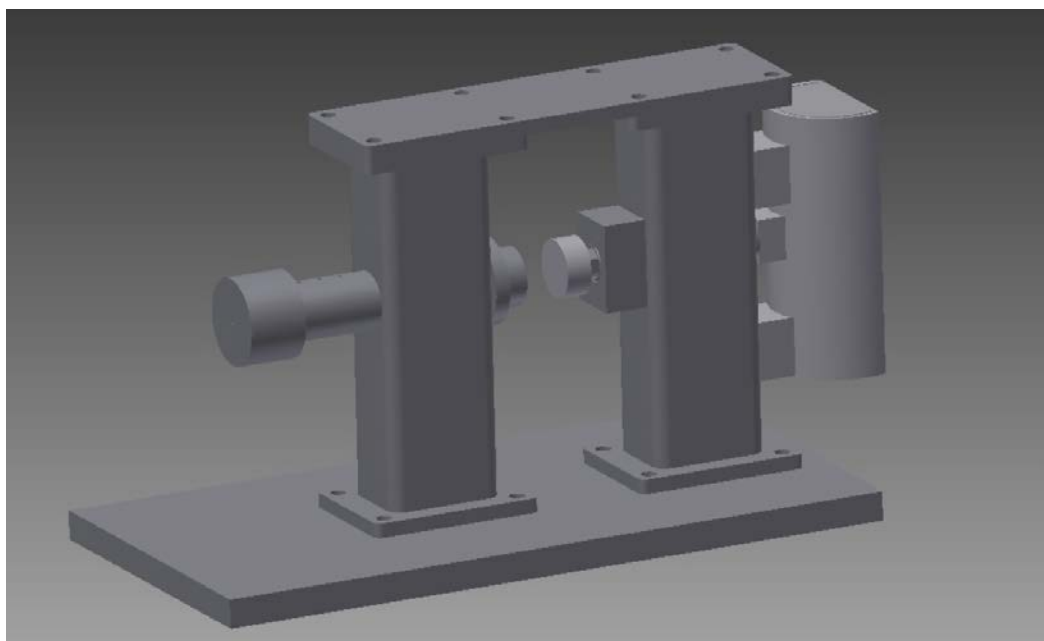
Significant drag phase acceleration would result in continued target acceleration throughout the positive phase. This result agrees with additional work performed by WRAIR researchers that demonstrated the importance of the diffraction phase of target-shock interaction in accelerating bluff bodies placed in the blast simulator.

Figure 4 is representative of the acceleration and velocity results of the blast exposed rats.

From this figure it can be seen that head is accelerated from rest to peak velocity during the diffraction phase of the exposure. In fact the acceleration occurs so quickly that peak velocity is developed within three frames after shock arrival. Three frames are the minimum number necessary to calculate an acceleration from position data. 25,000 frames per second is the limit of the laboratory video

recording equipment when the necessary resolution is preserved to allow the point tracking. These results agreed with similar findings from 2014 using video recorded at slower speed (5000 fps). Based upon these results, it was decided to treat the blast exposure and the resulting acceleration in the same manner as that which would

Fig 6: Impact device final design



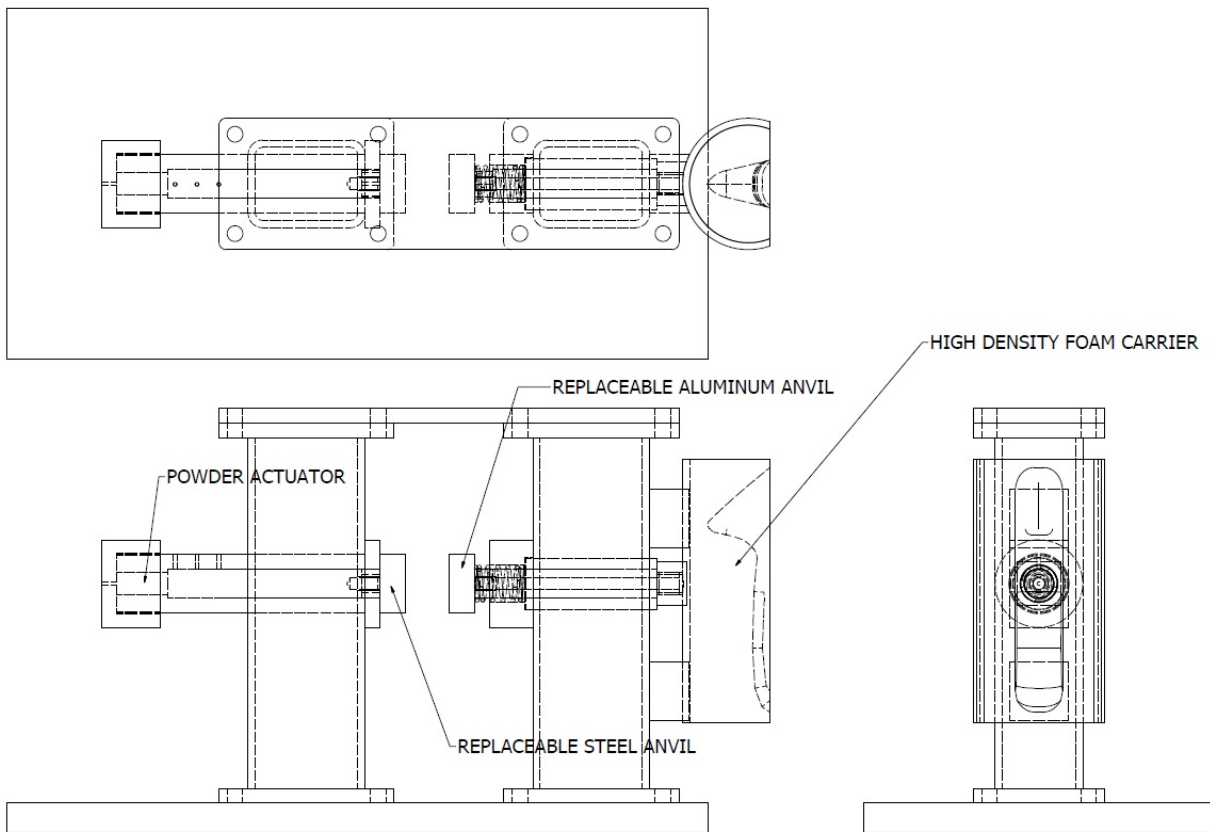
result from an impact exposure with the resulting force uniformly distributed over the projected surface of the target. The design focus of the acceleration device shifted from an attempt to recreate

a smooth acceleration path over several milliseconds to a method to uniformly distribute an impact loading over the projected surface of the target. Once this impact load is applied in less than 0.012 milliseconds, the target is allowed to follow a free flight path into a catch net located further down the ABS from the acceleration device.

Design and construction of acceleration test device:

The exact force required to accelerate the rat to the levels observed in the video is unknown. However a minimum required force can be estimated from the reflected shock pressure multiplied over the projected area of the rat. Assuming the rat has a projected area of 18 square inches and the exposure pressure is 20 psi incident, this will result in a direct reflected shock load of $2 \times 20 \text{ psi} \times 18 \text{ sq in} = 720 \text{ lbf}$. Higher loading force may be necessary. To apply this force, a robust foam lined carrier is needed. The weight of the carrier is several times that of the rat, so the device must be capable of developing impact force several times that of the above minimum. It was decided to develop an acceleration device that could produce an 8000 lb impact force applied over 0.5 inches in less than 0.2 milliseconds.

Fig 7: Impact device final design - split carrier shaft design



Three different methods were evaluated to develop this impact load: pneumatic, spring, and powder actuation. Practical pneumatic operators are limited to 150 psi operating pressure, requiring an operator 8 inches in diameter. The quick stroke necessary requires a specially designed and manufactured device. Springs that can produce 8000 lbf are very large (truck springs) and are not practical. A powder actuator consists of a chamber and barrel sufficient to contain the pressure resulting from firing a blank cartridge of smokeless gunpowder. Similar devices are in common use as drivers for nails in concrete and steel.

It was decided to develop a powder actuated device since it would be the most compact, simplest, and would allow the most potential for increased impact forces if necessary. The device could also be converted to a pneumatic operator if required, although this type operator would present more severe design and manufacturing challenges than a powder actuator.

A simple powder actuator was designed and constructed. This prototype device is shown in figure 5. In this prototype device the carrier plate is directly mounted to the captured bolt and the rat is positioned on the carrier plate using a dense foam bed that uniformly distributes the load force. A retaining ring is threaded onto the end of the barrel to prevent the bolt from exiting the barrel when fired. The device is fired using a 12 gauge shotgun blank.

When test fired, the device was demonstrated to be robust enough for safe operation, but the captured bolt was deformed by the retaining ring and resulted in an unrecoverable jam.

Consequently, an improved design was pursued to eliminate this possible failure mechanism. Figure 6 demonstrates the improved design.

In the final design the captured bolt is split and the carrier is installed on a separate shaft that is itself captured. The carrier travel is limited to 0.5 inches. The actuator bolt is allowed 2 inches total travel and impacts the carrier shaft after full pressure and velocity are developed. Actuator internal pressure is allowed to vent through three vent orifices located at the top of the actuator barrel. The breech is fully threaded onto the actuator barrel. The device is fired using an electric match pyrotechnic initiator.